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Serum Polychlorinated Biphenyls, Cytochrome P-450 1A1 Polymorphisms, and Risk of Breast Cancer in Connecticut Women

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Recent epidemiologic studies have suggested that genetic polymorphisms in the cytochrome P-450 1A1 gene (*CYP1A1*) may affect the relation between environmental exposure to polychlorinated biphenyls (PCBs) and breast cancer risk. The authors report results from a case-control study evaluating the potential effect of gene-environment interaction between *CYP1A1* and serum PCB levels on breast cancer risk among Caucasian women in Connecticut. The study included 374 case women with histologically confirmed breast cancer and 406 noncancerous controls with information on both serum PCB level and *CYP1A1* genotype (1999–2002). Compared with women who had the homozygous wild-type *CYP1A1 m2* genotype, significantly increased risks of breast cancer were found for women with the *CYP1A1 m2* variant genotype (odds ratio (OR) = 2.1, 95% confidence interval (CI): 1.1, 3.9), especially postmenopausal women (OR = 2.4, 95% CI: 1.1, 5.0). Risks associated with the *CYP1A1 m2* variant genotype were highest for all women (OR = 3.6, 95% CI: 1.5, 8.2) and postmenopausal women (OR = 4.3, 95% CI: 1.6, 12.0) with higher serum PCB levels (611–2,600 ng/g). The *CYP1A1 m1* and *m4* genotypes were not associated with breast cancer risk independently or in combination with PCB exposure. In summary, the *CYP1A1 m2* genetic polymorphism was associated with increased risk of female breast cancer and may modify the relation between PCB exposure and breast cancer risk.

breast neoplasms; cytochrome P-450 enzyme system; genetics; polychlorinated biphenyls; polymorphism (genetics); risk factors; women

Abbreviations: AHH, aryl hydrocarbon hydroxylase; CI, confidence interval; *CYP1A1*, cytochrome P-450 1A1; OR, odds ratio; PCB(s), polychlorinated biphenyl(s); PCR, polymerase chain reaction.

Polychlorinated biphenyls (PCBs) are strong inducers of key genes involved in steroid metabolism and xenobiotic metabolism, such as human cytochrome P-450 1A1 (*CYP1A1*) (1, 2). *CYP1A1* is a polymorphic gene involved in metabolism of steroids and several potentially genotoxic chemicals. So far, at least seven variant genotypes have been reported (3). Some epidemiologic studies have suggested a relation between *CYP1A1* genetic polymorphisms and risk of breast cancer, observing an increased risk (4, 5), while others have not found an association (6–8). Epidemiologic studies linking PCBs to breast cancer risk have also been inconsistent; although several earlier studies suggested a positive association (9–14), more recent studies showed no increased risk (15–17), with several even suggesting a negative association (18–21).

The lack of a consistent association between PCBs, *CYP1A1*, and breast cancer risk in earlier studies may suggest that only a portion of the study population, such as

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persons with specific *CYP1A1* variant genotypes, was susceptible to the adverse effects of PCB exposure. Indeed, two recent epidemiologic studies (22, 23) reported that women were at increased risk of breast cancer if they had a higher serum level of PCBs and a *CYP1A1 m2* genotype (exon 7 or isoleucine→valine), while independently neither factor was related to breast cancer risk. These observations suggest that *CYP1A1 m2* may modify the relation between PCB exposure and breast cancer risk. Toxicologic studies support these findings by showing that PCBs induce *CYP1A1* to metabolize environmental carcinogens into highly reactive intermediates, potentially resulting in DNA damage and ultimately carcinogenesis (24–26). Because of the widespread exposure to PCBs and the important role of *CYP1A1* in carcinogen activation and steroid hormone metabolism, we conducted a case-control study among Caucasian women in Connecticut to test the hypothesis raised by these earlier studies that women with higher levels of PCBs and specific *CYP1A1* genotypes have an increased risk of breast cancer.

MATERIALS AND METHODS

Study population

This study was built upon a recently completed case-control study of environmental exposure to organochlorines and female breast cancer risk in Connecticut. A detailed description of the parent study population has been previously published (16, 27). Briefly, 475 case women with histologically confirmed incident breast cancer (*International Classification of Diseases for Oncology* codes 174.0–174.9) and 502 randomly selected controls were identified in Tolland County and the New Haven County area, Connecticut, between January 1, 1994, and December 31, 1997. All cases and controls were aged 30–80 years, had no previous diagnosis of cancer (with the exception of nonmelanoma skin cancer), and were alive at the time of interview.

Potentially eligible cases and controls from the New Haven County area were identified using computerized patient information from Yale-New Haven Hospital (the major hospital in the area), where records of all newly completed breast-related surgeries are kept. We consecutively selected all breast cancer patients who met the study eligibility requirements as described above. A total of 326 cases with incident breast cancer were recruited. Efforts were made to frequency-match the cases and controls by age within 5-year intervals at a 1:1 ratio by adjusting the number of controls randomly selected in each age stratum every few months. A total of 347 controls were selected from the computerized files of women who had undergone breast-related surgery and were histologically confirmed to be without breast cancer or atypical hyperplasia. Controls included women diagnosed with benign breast diseases (including fibroadenoma, nonproliferative benign breast disease, and proliferative benign breast diseases without atypia) and women with normal tissue. For the subjects recruited from the New Haven County area, participation rates were 71 percent for controls and 77 percent for cases.

For Tolland County, we identified newly diagnosed breast cancer cases from Connecticut hospital records using the Rapid Case Ascertainment Shared Resource of the Yale Comprehensive Cancer Center. A total of 149 such case women were recruited. Population-based controls with Tolland County addresses were recruited using random digit dialing methods for persons below age 65 years and the Centers for Medicare and Medicaid Services for persons aged 65 years and above. A total of 155 such controls were recruited. Efforts were also made to frequency-match the cases and controls by age within 5-year intervals at a 1:1 ratio by adjusting the number of controls randomly selected in each age stratum. The participation rates in Tolland County were 61 percent for controls and 74 percent for cases.

After obtaining approval from each subject's hospital and physician, we approached potential participants by letter and then by telephone, and a trained interviewer interviewed those who consented, either at home or at a location convenient for the patient. A standardized, structured questionnaire was used to obtain information on major known or suspected confounding factors, including menstrual and reproductive history, lactation history, past medical history, family history of cancer (history of breast cancer and other cancers in first-degree relatives), occupation, diet, and demographic factors. Dietary information was collected using a scannable semiquantitative food frequency questionnaire developed by the Fred Hutchinson Cancer Research Center that was designed to optimize estimation of fat intake. Each subject was asked to characterize her usual diet during the year prior to being interviewed for our study.

Following the interview, the participant provided a blood sample, collected by venipuncture by study staff. The serum samples were used to determine PCB levels as described elsewhere (16, 27, 28), and the blood clots were used for *CYP1A1* genotyping. A total of 413 (87 percent) cases and 444 (88 percent) controls had information on both serum PCB level and *CYP1A1* genotype. Because the variant alleles in the *CYP1A1* gene vary in frequency among racial groups (8) and more than 90 percent of our subjects were Caucasians (with approximately 6 percent Blacks, 1 percent Asians, and 2 percent persons of other races), we restricted this analysis to Caucasians (374 cases and 406 controls).

The following PCB congeners were measured in this study: 74, 118, 138, 153, 156, 170, 180, 183, and 187. Serum residue results were determined in parts per billion on a wet-weight basis, which is equivalent to nanograms of compound per gram or milliliter of serum. The residues were also expressed on a lipid-adjusted basis as nanograms of compound per gram of lipid. Total PCB level was defined as the sum of levels of the nine measured PCB congeners.

CYP1A1 genotyping

High-molecular-weight genomic DNA was isolated for *CYP1A1* genotyping. DNA purity and yield were assessed by determining absorbances at 260 nm and 280 nm. Genotyping of *CYP1A1 m1*, *m2*, and *m4* was performed using a combination of polymerase chain reaction (PCR) and restriction fragment length polymorphism analysis. PCR reactions

contained 5 μl of Advantage 2 PCR buffer (Clontech, Palo Alto, California), 0.1 mM 2′deoxynucleotide-3′-triphosphate, and 1 μl of 50X Advantage 2 polymerase mix. All PCR plates contained a positive control for each of three polymorphisms and a negative control. Random samples were repeated to assure reproducibility.

The presence of the *CYP1A1 m1* polymorphism was determined with the PCR method previously described by Bailey et al. (6), using the primers 5′-GGCTGAGCAATCTGACCCTA-3′ and 5′-GGCCCCAACTACTCAGAGGCT-3′. Amplification conditions consisted of an initial denaturing step at 95°C for 1 minute, followed by 30 cycles of 30-second melting at 95°C and 1 minute of annealing at 68°C, followed by 1-minute extension at 68°C. The PCR product was digested with the restriction enzyme *Msp*I at 37°C for 1 hour and 45 minutes to yield either a 739-base-pair product in the absence of the polymorphism or two products (408 and 331 base pairs) in the presence of the polymorphism. The digestion products were electrophoresed in 3 percent agarose, stained with 0.05 percent ethidium bromide, and photographed under ultraviolet light.

The presence of the *CYP1A1 m2* polymorphism was determined with the PCR method previously described by Bailey et al. (6), using the primers 5′-GAAAGGCTGGGTCCACCCTCT-3′ and 5′-CCAGGAAGAGAAAGACCTCCCAGCGGGCCA-3′. Amplification conditions consisted of an initial denaturing step at 95°C for 1 minute, followed by 30 cycles of 30-second melting at 95°C and 3 minutes of annealing at 71°C, and then 1-minute extension at 71°C. The PCR product was digested with the restriction enzyme *Bsr*DI at 65°C for 1 hour and 80°C for 20 minutes to yield either two products (31 and 232 base pairs) in the absence of the polymorphism or one band (263 base pairs) in the presence of the polymorphism. The digestion products were electrophoresed in 1 percent low-melting agarose, stained with 0.05 percent ethidium bromide, and photographed under ultraviolet light.

The presence of the *CYP1A1 m4* polymorphism was determined with the PCR method previously described by Bailey et al. (6), using the primers 5′-GAAAGGCTGGGTCCACCCTCT-3′ and 5′-GGCCCCAACTACTCAGAGGCT-3′. Following denaturation at 95°C for 1 minute, 30 cycles of amplification were performed under the same conditions as those used for *m2*. The PCR product was digested with the restriction enzyme *Bsa*I at 50°C for 1 hour and 65°C for 20 minutes to yield either two products (1,199 and 494 base pairs) in the absence of the polymorphism or one band (1,693 base pairs) in the presence of the polymorphism. The digestion product was electrophoresed and stained as described for *m2*.

Data analysis

The *CYP1A1 m1*, *m2*, and *m4* polymorphisms were categorized as either wild-type (homozygous wild-type allele) or variant (heterozygous allele or homozygous variant allele). Serum levels of PCBs were categorized as high (higher than the median based on the distribution in the control group) or low (equal to or lower than the median based on the distribution in the control group).

Unconditional logistic regression was used to estimate the overall association between *CYP1A1* genotypes, serum PCB levels, and risk of breast cancer and to evaluate effect modification between PCB levels and specific *CYP1A1* genotypes with regard to breast cancer risk. The variables age (as a continuous variable), body mass index (weight (kg)/height $(m)^2$: <25, 25–29.99, or ≥30), lifetime duration of lactation (0, 1–5, 6–15, or >15 months), annual household income (tertiles based on the distribution in controls: <$9,200, $9,200–$18,400, or >$18,400), family history of breast cancer (history of breast cancer in a first-degree relative), menopausal status, and study site changed the odds ratios only slightly, but we kept these variables in the final model because of their recognition as confounders. Adjustment for other covariates, such as age at menarche, age at first full-term pregnancy, age at menopause, total fat intake, tobacco use, and alcohol consumption, did not show any effect on the odds ratios and thus were not included in the final model. Maximum likelihood estimates of the parameters were obtained using SAS (SAS Institute, Inc., Cary, North Carolina).

RESULTS

Table 1 presents the distribution of data on selected baseline characteristics for cases and controls. Compared with premenopausal women, postmenopausal women had a significantly increased risk of breast cancer, and women with later age at menopause also had a higher risk. Later age at first full-term pregnancy was associated with a slightly increased risk of breast cancer, though it was not statistically significant. While cases had a slightly higher fat intake, controls had a higher family income. No other baseline factors showed a material difference between cases and controls.

As table 2 shows, women with at least one *CYP1A1 m2* variant allele had a twofold increased risk of breast cancer (odds ratio (OR) = 2.1, 95 percent confidence interval (CI): 1.1, 3.9) compared with women with homozygous wild-type *CYP1A1 m2*, and the risk became greater among postmenopausal women (OR = 2.4, 95 percent CI: 1.1, 5.0). For women who carried *CYP1A1 m1* variant genotypes or *m4* variant genotypes, our data did not show any significantly increased risk of breast cancer in comparison with women who carried the *CYP1A1 m1* and *m4* wild-type genotypes, respectively. After stratifying the data by menopausal status, we still did not find any association with breast cancer risk for these two genotypes. The results were similar between the two study sites (data not shown). The lipid-adjusted serum level of PCBs in this study population ranged from 311 ng/g to 2,600 ng/g, with a median of 610 ng/g; this was comparable with most previous reports (14, 18, 21, 22). No significantly increased risk of breast cancer was associated with serum level of PCBs in this study.

Table 3 presents data on the association between *CYP1A1* genotype, serum PCB level, and breast cancer risk. We found a significantly increased risk of breast cancer for women who carried the *CYP1A1 m2* variant genotype and had a high serum PCB level in comparison with women with a low serum PCB level who carried the *CYP1A1 m2* wild-

TABLE 1. Selected characteristics of breast cancer cases and controls among Caucasian women in Connecticut, 1999–2002

Characteristic	No. of cases (n = 374)	No. of controls (n = 406)	OR*	95% CI*
Age (years) at menarche				
<11	29	27	1.0	
11–12	145	162	0.9	0.5, 1.6
≥12	200	217	1.0	0.6, 1.7
Menopausal status				
Premenopausal	85	145	1.0	
Postmenopausal	289	261	1.9	1.3, 2.6
Age (years) at menopause				
<44	72	82	1.0	
44–49	96	81	1.4	0.9, 2.2
>49	110	91	1.4	0.9, 2.2
Missing data	11	7		
Full-term pregnancy				
No	46	62	1.0	
Yes	328	344	1.1	0.7, 1.7
Age (years) at first full-term pregnancy				
<22	85	95	1.0	
22–26	135	135	1.2	0.8, 1.7
>26	108	114	1.3	0.9, 2.0
Lifetime duration of lactation (months)				
0	187	175	1.0	
1–5	45	59	0.7	0.5, 1.2
6–15	52	58	1.0	0.6, 1.6
>15	44	52	1.0	0.6, 1.5
History of breast cancer in a first-degree relative				
No	308	341	1.0	
Yes	66	65	1.1	0.8, 1.6
Body mass index†				
<25.0	203	240	1.0	
25.0–29.9	99	99	1.0	0.7, 1.5
≥30.0	72	67	1.1	0.7, 1.7
Fat intake (g/day)				
<44.4	106	132	1.0	
44.4–68.5	133	136	1.2	0.8, 1.7
>68.5	130	131	1.2	0.8, 1.7
Missing data	5	7		
Cigarette smoking				
Never	158	180	1.0	
Ever	216	226	1.0	0.8, 1.4
Alcohol consumption				
Never	49	54	1.0	
Ever	325	352	1.1	0.7, 1.7
Annual household income				
<$9,200	113	111	1.0	
$9,200–$18,400	109	118	0.9	0.6, 1.3
>$18,400	90	126	0.7	0.5, 1.0
Missing data	62	51		

* OR, odds ratio; CI, confidence interval.
† Weight (kg)/height (m)^2.

type genotype (OR = 3.6, 95 percent CI: 1.5, 8.2). This risk was higher among postmenopausal women (OR = 4.3, 95 percent CI: 1.6, 12.0). A borderline-significant increased risk of breast cancer was also found for all women and postmenopausal women with high serum PCB levels who carried *CYP1A1 m1* variant genotypes in comparison with women with a low serum PCB level who carried the *CYP1A1 m1* wild-type genotype (OR = 1.5 (95 percent CI: 1.0, 2.4) and OR = 1.8 (95 percent CI: 1.0, 3.2), respectively). No significantly increased risk of breast cancer was found among women who carried *CYP1A1 m4* genotypes with different serum levels of PCBs.

We had data on PCB levels in adipose tissue for 181 cases and 107 controls in the Yale-New Haven population. The results based on adipose tissue levels of PCBs showed a risk pattern similar to that based on serum levels of PCBs. Compared with women who had a low adipose tissue level of PCBs and carried wild-type *CYP1A1 m2*, postmenopausal women who had a high adipose tissue level of total PCBs and carried the *CYP1A1 m2* variant genotype had an odds ratio of 3.6 (95 percent CI: 0.4, 31.3). On the basis of PCB congeners' hormonal and nonhormonal activities, we found that postmenopausal women who had a high adipose tissue level of PCBs and carried the *CYP1A1 m2* variant genotype had odds ratios of 3.4 (95 percent CI: 0.4, 30.9) for congeners reported to have estrogenic activities (congener 187), 3.5 (95 percent CI: 0.4, 30.8) for congeners reported to have antiestrogenic activities (congeners 74, 118, 138, 156, and 170), and 4.9 (95 percent CI: 0.6, 43.3) for congeners reported to have genotoxic properties (congeners 153, 180, and 183; data not shown).

DISCUSSION

In this study, a twofold increased risk of breast cancer was found for women with *CYP1A1 m2* variants as compared with women with the *CYP1A1 m2* wild-type genotype, and a greater than fourfold increased risk was found for postmenopausal women with higher serum levels of PCBs and the *CYP1A1 m2* variant genotype as compared with those with a lower serum level of PCBs and the *CYP1A1 m2* wild-type genotype. A borderline-significant increased risk of breast cancer was found for women with the *CYP1A1 m1* variant genotype and higher serum levels of PCBs, but no increased risk was found for the *CYP1A1 m4* variant genotype, either alone or in combination with PCB exposure. We also observed that several factors showed the same risk pattern as recognized risk factors for breast cancer, such as age at first full-term pregnancy, age at menopause, and fat intake, though the findings were not statistically significant. Family breast cancer history was slightly associated with an increased risk of breast cancer in this study.

The potential effect modification between *CYP1A1 m2* genotype and PCB level in the risk of breast cancer observed among postmenopausal women in this study is consistent with the findings from two recent epidemiologic studies (22, 23). A case-control study from Upstate New York (23) examined the relation between serum levels of PCBs, *CYP1A1* genotypes, and breast cancer risk among 154 breast cancer cases and 192 controls. Moysich et al. (23) reported

TABLE 2. Relations of *CYP1A1* genotype and serum level of polychlorinated biphenyls to risk of breast cancer, overall and by menopausal status, among Caucasian women in Connecticut, 1999–2002*

	Total				Premenopausal women				Postmenopausal women			
	No. of cases	No. of controls	OR†	95% CI†	No. of cases	No. of controls	OR	95% CI	No. of cases	No. of controls	OR	95% CI
CYP1A1 genotype												
m1												
WT/WT‡	279	324	1.0		68	115	1.0		211	209	1.0	
Variants§	95	82	1.1	0.7, 1.6	17	30	0.8	0.4, 1.8	78	52	1.2	0.7, 1.8
m2												
WT/WT	334	385	1.0		79	137	1.0		255	248	1.0	
Variants	40	21	2.1	1.1, 3.9	6	8	1.4	0.4, 5.1	34	13	2.4	1.1, 5.0
m4												
WT/WT	346	369	1.0		80	131	1.0		266	238	1.0	
Variants	28	37	0.8	0.5, 1.4	5	14	0.6	0.2, 1.8	23	23	0.9	0.5, 1.7
Serum level of polychlorinated biphenyls (ng/g)												
Low (310–610)	173	202	1.0		30	60	1.0		143	142	1.0	
High (611–2,600)	201	204	1.2	0.9, 1.6	55	85	1.3	0.7, 2.3	146	119	1.2	0.9, 1.7

* Odds ratios were adjusted for age (as a continuous variable), body mass index (weight (kg)/height $(m)^2$: <25, 25–29.99, or >29.99), lifetime duration of breastfeeding (never or 1–5, 6–15, or >15 months), history of breast cancer in a first-degree relative, menopausal status, annual household income (in tertiles based on the distribution in controls), study site, and genotype (three genotypes, adjusted for each other).

† OR, odds ratio; CI, confidence interval.

‡ Homozygous wide-type alleles.

§ One variant allele or homozygous variant alleles.

an odds ratio of 3.0 (95 percent CI: 1.2, 7.5) among postmenopausal women with serum PCB levels above the median and at least one variant allele as compared with women who had lower PCB levels and were homozygous for the isoleucine allele. In the same study, Ambrosone et al. (5) reported an odds ratio of 1.6 (95 percent CI: 0.9, 2.8) for women with *CYP1A1 m2* variants as compared with women who were homozygous for wild-type *CYP1A1 m2*.

An increased risk of breast cancer among postmenopausal women with higher body levels of PCBs and specific *CYP1A1* variants was also reported by Laden et al. (22) in a nested case-control study involving 367 breast cancer cases and an equal number of controls from the Nurses' Health Study cohort. Laden et al. found that while there was no independent association of either *CYP1A1* variants or PCB levels with breast cancer risk, a relative risk of 2.8 (95 percent CI: 1.0, 7.8) was observed among postmenopausal women with plasma PCB levels in the highest tertile and *CYP1A1 m2* variants, as compared with women who were homozygous for wild-type *CYP1A1 m2* and had PCB levels in the lowest tertile.

Potential effect modification between PCB level and a specific *CYP1A1* genotype is biologically plausible. PCBs are potent *CYP1A1* inducers, and *CYP1A1* is involved in hormone metabolism. The *CYP1A1* locus encodes aryl hydrocarbon hydroxylase (AHH), also called 17-β-estradiol (E2) hydroxylase, which is involved in steroid hormone metabolism. AHH catalyzes the monooxygenation of polycyclic aromatic hydrocarbons to phenolic products and epoxides that are mutagenic and carcinogenic (25). For example, AHH catalyzes the first step in the activation of benzo(*a*)pyrene to its mutagenic form, which can produce mutagenic DNA adducts in breast tissue (29). The role of AHH in both carcinogen activation and estrogen metabolism suggests that the PCB-mediated enhanced *CYP1A1* activity could lead to an increased risk of breast cancer.

In our study, a significantly increased risk of breast cancer was found only for the *CYP1A1 m2* genotype. Earlier experimental studies appeared to support the hypothesis that the *CYP1A1 m2* genotype is more likely to be related to breast cancer risk (24, 26). It has been shown that *CYP1A1* activity is more readily inducible in persons with the *m2* genotype as compared with other *CYP1A1* variant genotypes (26). Greater inducibility would result in higher levels of the protein, leading in turn to greater carcinogen bioactivation and therefore a higher potential risk of cancer (24, 26).

In interpreting results from the current study, several potential limitations should be considered. One is that we included patients with benign breast disease in the control group in New Haven County and in a population-based control group in Tolland County. However, the lack of association for serum PCB levels and breast cancer is unlikely to be entirely explained by the inclusion of benign breast disease patients as controls. In the multivariate analysis carried out by study site, we reached the same conclusions as in the combined analysis. In addition, two previous studies using benign breast disease patients as controls reported a positive association between PCBs and female breast cancer risk (30, 31).

Another potential concern is that using serum levels of PCBs rather than adipose tissue levels may not reflect the real body burden of PCBs. However, studies have shown that there is good correlation between lipid-adjusted serum levels of PCBs and adipose tissue levels of PCBs (32). In

TABLE 3. Risk of breast cancer according to *CYP1A1* genotype and serum level of polychlorinated biphenyls, overall and by menopausal status, among Caucasian women in Connecticut, 1999–2002*

CYP1A1 genotype	Serum level† of polychlorinated biphenyls	Total				Premenopausal women				Postmenopausal women			
		No. of cases	No. of controls	OR‡	95% CI‡	No. of cases	No. of controls	OR	95% CI	No. of cases	No. of controls	OR	95% CI
m1													
WT/WT‡,§	Low	132	166	1.0		23	51	1.0		109	115	1.0	
Variants¶	Low	41	36	1.5	0.9, 2.4	7	9	1.6	0.5, 5.1	34	27	1.4	0.8, 2.5
WT/WT	High	147	158	1.3	0.9, 1.8	45	64	1.6	0.8, 3.0	102	94	1.1	0.8, 1.7
Variants	High	54	46	1.5	1.0, 2.4	10	21	1.0	0.4, 2.6	44	25	1.8	1.0, 3.2
m2													
WT/WT	Low	157	189	1.0		27	55	1.0		130	134	1.0	
Variants	Low	16	13	1.6	0.7, 3.5	3	5	1.1	0.2, 5.0	13	8	1.8	0.7, 4.5
WT/WT	High	177	196	1.2	0.9, 1.6	52	82	1.3	0.7, 2.3	125	114	1.1	0.8, 1.6
Variants	High	24	8	3.6	1.5, 8.2	3	3	2.2	0.4, 1.2	21	5	4.3	1.6, 1.2
m4													
WT/WT	Low	161	183	1.0		28	56	1.0		133	127	1.0	
Variants	Low	12	19	0.7	0.3, 1.5	2	4	1.0	0.2, 6.0	10	15	0.6	0.3, 1.5
WT/WT	High	185	186	1.2	0.9, 1.6	52	75	1.4	0.8, 2.5	133	111	1.1	0.8, 1.6
Variants	High	16	18	1.1	0.5, 2.2	3	10	0.6	0.1, 2.5	13	8	1.7	0.7, 4.2

* Odds ratios were adjusted for age (as a continuous variable), body mass index (weight (kg)/height $(m)^2$: <25, 25–29.99, or >29.99), lifetime duration of breastfeeding (never or 1–5, 6–15, or >15 months), history of breast cancer in a first-degree relative, menopausal status, annual household income (in tertiles based on the distribution in controls), and study site.

† Low, 310–610 ng/g; high, 611–2,600 ng/g.

‡ OR, odds ratio; CI, confidence interval; WT, wild-type.

§ Homozygous wide-type alleles.

¶ One variant allele or homozygous variant alleles.

addition, our study results based on adipose tissue PCB levels showed a risk pattern similar to that based on serum PCB levels. PCBs represent a heterogeneous group of 209 possible congeners, with some displaying estrogenic properties, some eliciting antiestrogenic activity, and some exhibiting genotoxic properties (12). Epidemiologic studies linking individual congeners to breast cancer risk have yielded conflicting results (12, 21, 27, 33). For our small number of subjects with data on adipose tissue levels of PCBs, we did attempt to analyze the relation by PCB congener; those results appeared to suggest that the risk of breast cancer associated with PCBs and *CYP1A1* genotypes may vary by individual or group congeners of PCBs. Thus, the effect of gene-environment interaction between *CYP1A1* genotype and individual or group congeners of PCBs on breast cancer risk merits further investigation.

Unlike prospective studies, in which researchers can collect biologic samples long before disease onset and from different periods of human development, case-control studies like this one rely on biologic samples collected after disease onset. One limitation of this study design is that current levels of PCBs may not represent the levels subjects had when their disease developed—especially since the manufacture and use of these compounds has been banned in the United States since the late 1980s.

Although we had 374 incident breast cancer cases and 406 controls, the relatively low prevalence of the variant genotypes limited our statistical power to stratify the data by major confounders, such as lactation and menopausal status. Larger population-based studies are warranted to confirm these findings and to investigate whether this interaction between *CYP1A1 m2* and PCBs is modified by these factors.

The prevalence of the *m2* genotype was 6 percent in our control group. In previous studies, the prevalence of this genotype in Caucasian women ranged from 7 percent to 17 percent (4–8, 22, 23). Thus, our values are consistent with those of previous studies, though at the low end of the range. It is interesting to note that for those studies reporting the specific population considered, all of the studies with *m2* prevalences at the high end of the range (12–17 percent) investigated populations in Upstate New York (4, 5, 23), while those with prevalences at the low end of the range (6–7 percent) considered populations elsewhere (our study and the studies by Bailey et al. (6) and Rebbeck et al. (8)). Considering all of the studies, it appears that this polymorphism occurs with a relatively high frequency in the general population; this has important public health implications.

In summary, recent epidemiologic studies have suggested potential effect modification between body levels of PCBs, the *CYP1A1 m2* genetic polymorphism, and breast cancer risk. The results are biologically plausible, because PCBs are potent *CYP1A1* inducers, and the *CYP1A1 m2* genotype is more readily inducible than other *CYP1A1* genotypes. PCB-mediated enhanced *CYP1A1 m2* activity would produce greater levels of bioactivated carcinogens than would other *CYP1A1* genotypes. Considering the widespread exposure to PCBs and the relative commonness of the high-risk genotype in the general population, it is essential to replicate these findings in different populations with larger sample sizes.

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